MOLD GROWTH ON WAFERED HAY

by

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INTRODUCTION

Through the centuries man has had various needs that had to be satisfied; and man has usually had the capabilities to study each need and to come up with a solution that would satisfy it. History clearly points out that where the need and capability were matched, progress was achieved. The first attempt towards progress made by man was the domestication of animals. Later, the steam engine, electric lights, automobiles, and presently the race for space are all good examples of what man has done to satisfy various needs.

During the winter season in temperate climates when there is no place for the animals to graze, man has had to assume the responsibility of providing food for domestic animals. This has meant much time and back-breaking labor during the summer season storing animal food for the winter.

Much time and thought towards reducing this labor have been devoted by research engineers, economists, extension personnel, and particularly the people directly concerned - the farmers. Methods for supplying water and grain to cattle have been greatly improved to the point where little labor and attention are required.

However, the supply of roughage, particularly alfalfa hay, still requires much manual labor. The packaging of hay in the field for efficient storage, and its removal from storage for distribution to livestock, is one of the most difficult operations to completely mechanize.

Hay is one of the most important feeds for livestock, and the annual production of hay in the United States averages over 100 million tons according to Barger (4). A possible aid to the mechanization of hay handling, would be to form the hay into small, dense, regularly shaped packages. The

hay wafering machine does this by picking up the hay from the windrows and compressing it into small dense packages called wafers.

The term "Hay Wafer" for the purposes of this thesis, and, as defined by the Hay Pelleting Committee of the American Society of Agricultural Engineers (19) means: "An agglomerated unground forage which has some fibers equal to or greater than the length of its minimum cross section dimension". This agglomeration is packed into packages with or without the use of an artificial binding agent. The term "Wafer" is different from the term pellet. Roughage loses its identity by undergoing a grinding process before being pelleted.

The packaging of hay in wafer form has resulted in a storage problem. Prevention of mold seems to be the most serious problem in preserving quality while the wafers are in storage. This is the problem to be solved in this thesis. It is desired to learn how long does it take for mold to appear after the wafer is made. This will be accomplished by storing wafers in an environmental chamber, where conditions will be controlled. Visual observation will determine the time when mold first appears.

PURPOSE

As part of a larger field of study by the Agricultural Engineering Department at Kansas State University, the objective of this investigation was to determine how long after the wafer is made and stored under certain environmental conditions, does it take for mold to appear.

REVIEW OF LITERATURE

Hay Quality Determination

According to Gilbert (6) there is general disagreement by people directly concerned as to what should be used as an indicator of hay quality. Meyer (11) says that the ultimate test of hay quality is the animal response to growth, fattening or milk production; however, a simple chemical analysis can give some indication of the nutritive value. Liener (10) says we must keep in mind the protein efficiency; that is to say, the increase in body weight per unit quantity of protein. By heating the product to a temperature above 140°F, reduction per unit quantity of protein consumed was found. This is presumably due to non-enzymatic browning. Protein heated in the presence of carbohydrates is modified by the interaction of the free amino groups and the reducing sugars. He goes on to say, that if a chemical analysis is run, the total amount of protein present will be equal to that in the hay that has not been heated.

Storage Losses

Haelain et al. (7) say that wafers retain vitamin A better than loose hay or baled hay after drying and storage. This may be attributed to the exclusion of air from the wafer. According to Meyer (11), hay is known to lose carotene quite rapidly, but wafering may slow the process. Fulton (5) indicated that the preservation of pellets in an inert gas enhanced the quality due to the gas passing more readily through the mass to displace oxygen. Experience in Kansas by Reece et al. (14) showed that wafers in the range of 15 to 25% moisture wet basis, molded heavily over the surfaces on piles of wafered hay. Sometimes mold would be found next to the floor

of the structure, particularly so if the floor were concrete. The surface mold seemed to disappear after a period of time.

It is believed that wafers mold because the moisture content exceeds the critical 15% level. Also, it is difficult to get air circulation through the wafer because of its density. This problem is not evident in loose or baled hay, because the moisture content is usually below the critical level and the density is low enough to allow natural drying.

Wait (20) conducted mold growth experiments and found mold first occured at the cut ends and the nodes of the hay. The presumed reason was that the hyphae did not have to penetrate the outer layer to find food. The ends had plenty of available food.

Snow et al. (18) found that the main factors affecting mold growth are:

1. The relative humidity rather than the moisture content. After two years of storage at a relative humidity below 65%, no mold was present.

2. The temperature of storage.

3. Type of mold species present. At high relative humidity all species grew fast, but at low relative humidities only a few species were able to grow.

Snow (17) in a subsequent investigation found that between relative humidities of 90 - 100% all species of mold grew; below 90%, members of the Mucorales and fungi-imperfecti were isolated; below 75%, Penicillium spp. Aspergillus spp were able to develop under restricted moisture supply. Aspergillus repens and ruber were the most damaging to food stuffs.

According to the March, 1965, Special report from the Agricultural Research Service (1), scientists generally are inclined to conclude that

mold poisoning among livestock and poultry in this country is relatively infrequent and sporadic. However, they emphasize that extensive research has not been done in this field.

Precise information on the variables that cause certain common molds to produce toxic byproducts are not known. Consequently, mold prevention is receiving more emphasis as a practical means of avoiding the possible danger of mycotoxins. Bad odors, loss of seed viability, and the loss of palatability and nutritional value are also good reasons for the prevention of mold growth.

Myotoxins are toxic products from common molds. For example, a common mold like Apergillus flavus was found in peanut meal, and was found to have a toxic byproduct which was given the name of Aflatoxin.

In the handling and harvesting of products, proper measures should be taken to prevent mold from attacking the product. Modern farming techniques make it possible to keep mold to a minimum. But, with mechanization, crops are harvested at higher moisture contents. On the other hand, artificial drying has made the farmer more independant of the weather. Thus, the farmer is able to produce top quality products at the peak of their nutritive value, even in damp weather.

The recommended procedures, according to the March, 1965, Report of the Agricultural Research Service, for preventing mold damage vary with the crop. The moisture content is the single most important factor contributing to mold growth. Warm temperatures (60 to 100^{o}F) in combination with high relative humidities encourage mold growth.

It is also possible to get mold contamination from irrigation, and dormant mold in seeds.

According to A. M. Altschul (2), the digestibility of the leaf protein is reported to be reduced about one-half by drying, due to the use of high temperature drying air. Feeding trials on rabbits and chickens revealed that the animals fed on alfalfa meal dried by low temperature drying air, grew about twice as fast as the animals fed on meal dried by high temperature air. The author was emphatic to point out that a chemical analysis was run on both feeds and the two were found to have identical percentages of protein. Yet, there was a marked difference in the growth rate of the animals.

Jefferson (9) says that the greatest potential value of the properties of ionizing radiation lies in food processing. The extent of the control of microorganisms will depend on the dose rate and the environmental conditions employed after treatment. Long term storage can only be achieved by sterilization, which in this context means the practical elimination of all microorganisms. Enzymatic, chemical and physical changes in the food must also be controlled. In soft fruits, doses of 0.1 and 0.5 M rad were used to control mold activity. The incidence of mold attack on strawberries was markedly reduced during storage in refrigeration and at high temperatures. The length of time the storage is prolonged is dependant on the dose given. At low doses mycelia are still viable.

There is some loss in vitamins due to radiation, but no serious losses are caused to the nutritive value of the macronutrients, even at radiation sterilization doses. However, some losses of the biological value of protein have been noted.

The surgeon general's office said that little or no evidence of toxicity in irradiated foods was present.

EQUIPMENT AND METHOD

Equipment

The machine used to produce the wafers for this project was designed and built by Mr. F. N. Reece, (see plate I, page 9) Assistant Professor, Department of Agricultural Engineering at Kansas State University. The wafers were formed by a closed die process (15). The hay was forced into a cavity, or closed end die, held under pressure until the wafer was formed, and then ejected from the die or cavity. Forming pressures were approximately 2000 pounds per square inch, and the hold time varied from 13 seconds to 28 seconds depending on the moisture contents. The higher the moisture content, the longer the hold time.

A walk-in type Army surplus refrigerator was equipped with a duct system, which included a set of heating elements. The heating elements were capable of producing 6,000 watts which would be the equivalent of 341 BTU/min of heat. A fan was installed in the duct system which would deliver 1000 cubic feet per minute. The air was recirculated from the room to and through the duct system, and back into the room. The temperature of the room was controlled with a highly snesitive thermostat which controlled the heaters. Also, a sensitive thermostat was set on the refrigeration system to aid in keeping the temperature as constant as possible.

A Toledo 30-pound capacity balance scale, which could be read to 0.01 pounds, was used to weigh the samples before and after they were oven dried.

Thirty-two one-gallon jars were used to store the wafers in the room. Two copper tubes were installed on the lids of each jar. One reached the bottom of the jar, and through it air was pumped at a rate of 0.025 cubic

EXPLANATION OF PLATE I

A view of wafering machine in center of picture. Mr. F. N. Reece is the designer and builder of the wafering machine. He shows how the unheated hay was placed in the wafering machine. To the left, is the recording unit for temperatures. To the right, is the power unit for the hydraulic system for the wafering machine.

PLATE I

feet per hour. The other was much shorter, and was used as an exhaust for the incoming air, the location of a passage way for the thermocouple wire and, for the sampling of the carbon dioxide content.

As a safety device, the author built a bubble trap (see plate II, page 12) through which the air being pumped to the jars must pass. This aided in regulating the flow, and also, gave an indication of any restriction of air flow. Air was brought into the room from the main compressor unit in the research laboratory, and run through a 50-foot roll of copper tubing. The assumption was that the air would have a chance to come to equilibrium with the temperature of the room before it entered the jars. A pressure regulator was used to reduce the pressure from 90 to 30 pounds per square inch gauge. After leaving the pressure regulator, the air passed through a flowmeter used to measure the volume of air flow. From the flowmeter, the air proceeded through the manifold to the bubble traps, and then to the jars.

Copper-constantan thermocouples in conjunction with a Minneapolis-Honeywell Brown recording potentiometer which was controlled by a time switch and stepping switch, were used to record the temperature in the jars every hour.

A Foxboro Dew Cell element was used in conjunction with a portable potentiometer, to measure the relative humidities in the jars. A hose pump extracted the air from the jars which contained the wafers, and pumped it to a jar with the dew cell element in it. A reading was taken from the potentiometer, and from calibration tables the dew point was determined. With this figure, and the temperature of the room, the relative humidity was determined from a psychrometric chart.

EXPLANATION OF PLATE II

A view of the inside of the environmental chamber showing the flowmeter, manifold, and bubble trap used for the distribution of air to the jars. The jars with the air hoses and thermocouple installed are also shown.

PLATE II

A Kitagawa Precision Gas Detector in conjunction with detector tubes, was used to determine the carbon dioxide content in the jars. A vacuum pump was used to pull a 100 cubic centimeter sample of air through the detector tubes. The presence of carbon dioxide was detected by discoloration of the material inside the tubes. The tubes were then placed over calibration charts to determine the amount of carbon dioxide.

One phase of the study involved irradiation of some of the wafers to determine the effect of radiation on mold growth. A Gammacell 22 unit, belonging to the Department of Nuclear Engineering, (see plate III, page 15) was used to irradiate 25% moisture hay wafers. The radiation source was 12 type C 167 Pencils containing Aluminum Clad Cobalt 60 Slugs. On March 16, 1965, the measured dose rate of the Gammacell 22 was 3.27×10^5 Rads/Hr ± 5.1%. The total activity was 3963 curies.

The general overall procedure of the investigation will be briefly discussed in the following paragraphs.

Procedure

The alfalfa used was prepared from bales that were stored at the Agronomy farm of Kansas State University. The hay was chopped to lengths of 1 to 3 inches by a hammer mill. After the moisture content of the baled hay was determined, enough water was added to the chopped sample to bring the hay up to the desired moisture content for the experiment. Next, the samples were mixed in a mixer, and stored in sealed containers for a period of 48 hours at a temperature of 40° to 45° F. This was intended to give a more uniform moisture content throughout the sample. After the wafers were made, they were stored in gallon jars in a room at a temperature of

EXPLANATION OF PLATE III

A view of wafers in the plastic sandwich bags placed in the Gammacell 22 before being exposed to Gamma radiation.

PLATE III

90° F. Oven drying procedures were used to determine the moisture contents.

The hay, after being held in a refrigerated environment for 48 hours, was mixed by a mechanical mixer as shown in plage IV, page 18. Samples of 3/4 of a pound each were taken from the mixer and placed in the wafering machine as shown in plate I, page 9. Two different types of wafers were made: some were made from hay that was at ambient temperature, and the rest from heated hay. Heating the hay was accomplished by using an auger to convey the hay under infra-red heat lamps. The temperature of the heated hay varied from 135 to 155° F. Four different moisture contents were used; for unheated hay, 15, 20, 25, and 30% initial moisture content hay was used. For the heated portion of the experiment, hay of 20, 25, 30 and 35% initial moisture content was used. The reason for using the 5% increase in moisture on the heated wafers was that it was hoped that about 5% would be lost due to heating.

A moisture check revealed that the heated wafers with 20% moisture lost from 8.5 to 9.0%; the 25% wafers from 4.5 to 5.0%, and the 30% wafers lost from 3.3 to 3.8%. It was not possible to obtain this check on the 15 and 35% moisture wafers. They were mixed in single batches. The 20%, 25%, and 30% moisture samples were prepared together; half for the unheated experiment, and the other half for the heated experiment. The amount of moisture lost due to heating was determined from the results.

The unheated samples of hay were taken directly from the mixer and placed in the wafering machine, because the auger on the wafering machine would not convey the unheated hay properly. The heated samples of hay were conveyed by the auger under the heat lamps on their way to the closed die wafer forming chamber.

EXPLANATION OF PLATE IV

A view of the mixer used for mixing hay before being placed in the wafering machine. Beside it is the can in which the hay was stored at 45° F.

PLATE IV

The wafers were collected and placed in one-gallon jars in the refrigeration room. Lids were placed on the jars, the air hose placed on the longer of the copper tubes, thermocouples inserted as shown in plate II, page 12, and the room temperature brought to 90° F. Relative humidity readings were taken at approximately six hour intervals. A Brown recorder was used to record the temperatures in the jars every hour. Visual observation was made periodically to check the progress of the mold growth.

Air at room temperature was pumped through the jars. This assured proper environment and enough oxygen for mold to grow, and yet did not alter the moisture content of the wafers by drying due to the air coming in.

The weight of the jars as shown on plate V, page 21, was recorded before, and at the end of each experiment, or when mold was visible. This enabled the author to check for any changes that may have taken place during the experiment, such as loss of moisture. Once the appearance of mold had been detected, a final relative humidity reading was taken. The wafers were then removed from the jars and placed in the drying ovens for a period of 48 hours at 100° C. All moisture percentages were expressed on a wet basis, as recommended by the Hay Pelleting Committee of the American Society of Agricultural Engineering.

At the end of each experiment, all of the tubes, lids, jars, and thermocouple wires were disinfected with Roccal Sanitizing (16) solution at a strength of 800 parts per million.

The die used in making the heated wafers was kept at a temperature of about 45° F. by circulation of water through it. The cold die and the heated hay tended to produce a hard layer around the outer surface of the wafer leaving the interior still soft and spongy. It would appear that the harden-

EXPLANATION OF PLATE V

Wafers in the one-gallon jars with lid and copper tubes. The jars were weighed before and after storage to be sure that no loss of moisture was occuring.

PLATE V

ing of the outer surface might give the wafer increased durability and less loss of fines during handling, yet produce a wafer soft enough for the cattle to eat easily.

The 25% moisture wafers that were used for the radiation experiment, were put in plastic sandwich bags, and placed in the Gammacell as shown in plate III, page 15. They were exposed to a dose of 320,000 roentgens ± 20%. After being irradiated they were taken out of the bags, placed in the one gallon jars, and handled in the same manner as all of the other samples.

It should be pointed out at this time that the radiation used consists of two photons that leave Cobalt 60 sources with a short wavelength and high energy (1.17 and 1.33 Mev). These bundles of energy travel at the speed of light, and pass right through the material being irradiated. For this radiation to be effective in destroying mold, the gamma ray as it passes through the mold spore, must react with the spore; this reaction will cause the necessary changes for the spore to die. If the gamma ray goes through the mold spore without reacting, then the spore is still viable.

No statistical analysis was needed, because it was evident that the data obtained would provide highly significant results for any test run, at any rejection level. Professor A. M. Feyerherm, from the Department of Statistics at Kansas State University, was consulted on the matter.

RESULTS AND CONCLUSIONS

The wafers were 2 X 2 1/2 inches in cross section, and lengths varied from 3 1/2 to 5 1/2 inches, depending on the feed rate of the auger and the moisture content of the hay in the case of the heated hay, and on only the moisture content in the case of the unheated wafers. As the moisture content

increased, and after the wafer was ejected from the die, they would expand anywhere from 1/8th of an inch to 3/4th of an inch in all directions. The wafers made from the 15, 20, and 25% moisture hay had good appearance, kept their shape, and expanded very little. The wafers made from the 30 and 35% moisture hay had little or no similarity to a wafer. They looked more like a mass of hay with no shape and were very wet, soggy and sticky.

By observing plates VI, and VII, pages 25 and 27, it can be seen that there is a definite relationship between the moisture content of the wafer, and the time for mold growth to become visible. It must be pointed out that any time the temperature is between 59^{o} F. and 131^{o} F., with a relative humidity above 65%, the mold development starts. These environmental conditions activate the enzymes in the mold spore. Next, the hyphae come out of the spore much like the root and leaf from a seed, and penetrate the alfalfa stems and leaves to obtain the food necessary for their development. As time goes by, a white fuzzy material becomes visible. According to Dr. C. Kramer, of the Botany Department at Kansas State University, this is known as the vegetative growth. The mold continues to grow by the decomposition of the plant structure. Later, green and blue dots appear among the white fuzzy growth and this stage of development, according to Dr. C. Kramer, is called sporulation. Actually, it is new mold spore. From these blue and green dots more white fuzzy vegetative growth develops, with the hyphae again penetrating the plant to get their food for continued development. This process is repeated over and over again.

For the purposes of this experiment, it was assumed by the author that the conditions for mold to grow were present soon after the wafers were

EXPLANATION OF PLATE VI

Effect of moisture content on the time for mold to grow on unheated wafers.

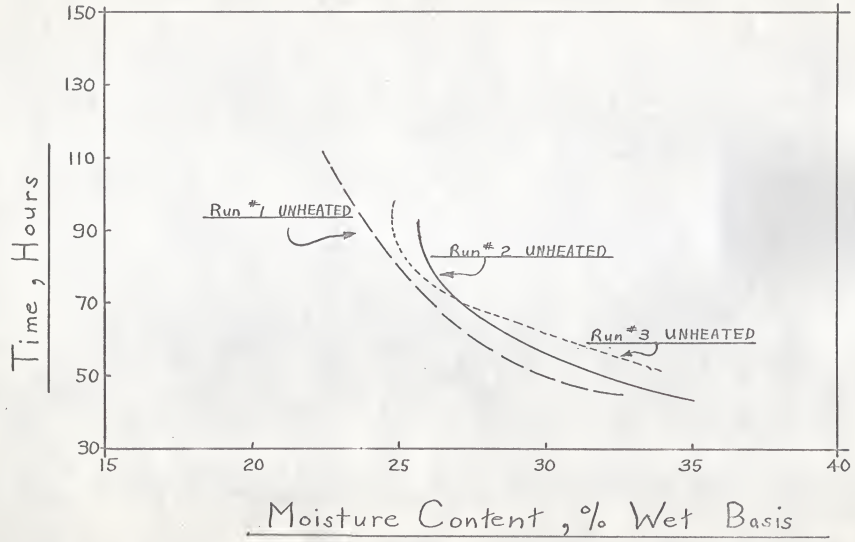

150
130
110
90
70
50
30
Time, Hours
Run #1 UNHEATED
Run #2 UNHEATED
Run #3 UNHEATED
15
20
25
30
35
40
Moisture Content, % Wet Basis

PLATE VI

EXPLANATION OF PLATE VII

Effect of moisture content on the time for mold to grow on heated wafers.

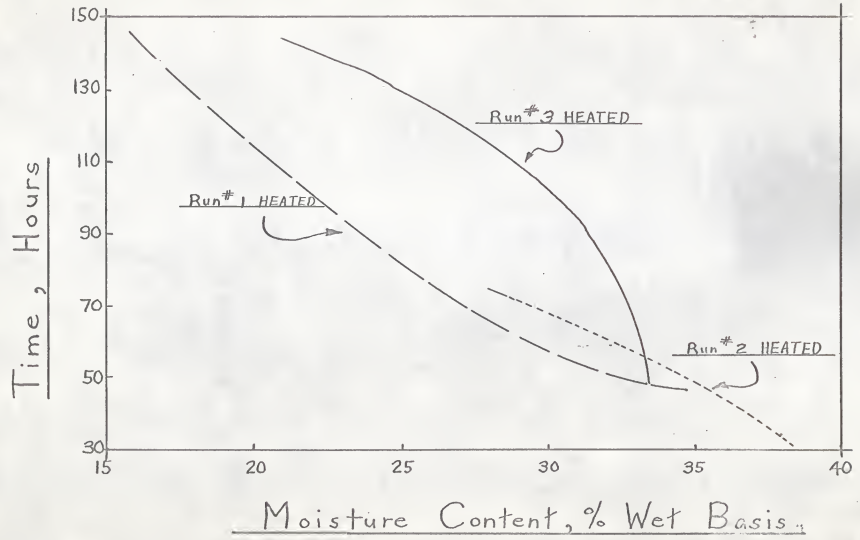

Time, Hours
150
130
110
90
70
50
30
15
20
25
30
35
40
Run # 3 HEATED
Run # 1 HEATED
Run # 2 HEATED
Moisture Content, % Wet Basis

PLATE VII

placed in the jars. The time started as soon as the wafers were placed in the jars, and terminated at the first evidence of mold growth. Usually, this was detected by a rise in temperature recorded by the thermocouple installed in the jar, as shown on plate VIII, page 30. After seeing this rise in temperature visual inspection of the jars would detect the presence of mold as shown in plate IX, page 32. In this way, the author was able to safely record the exact time, within an hour, at which the mold became visible. It can be definitely said that temperature recordings by the use of the thermocouple can be used as a means of detecting mold growth.

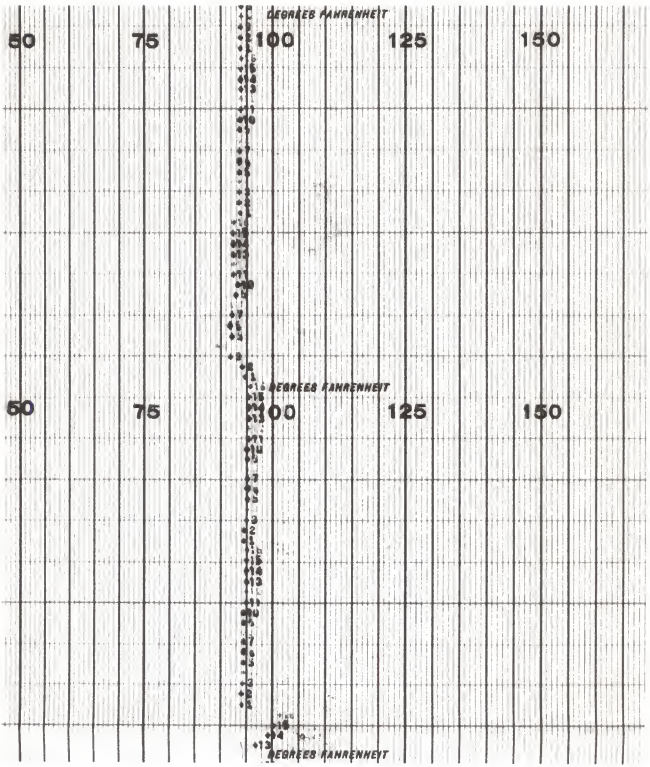
A closer examination of plate VI, page 25, shows that for unheated wafer samples, the curves at the lower end would seem to project asymptotically along a line near the 30 to 36 hour mark. This would be logical because at least 24 to 36 hours are required for mold to grow under optimum conditions (3). Obviously we did not have optimum conditions, but must have been reasonably close to them and should have obtained similar results.

The other end of the curve also projects asymptotically towards infinity. This does not seem to be too realistic for several reasons. If mold growth depends upon moisture content of the hay only, a curve approaching infinity would be correct. However, the temperature and relative humidity must play an important role in mold growth, as evidenced by the constant change in relative humidity during the experiment. The moisture contents reported are the average moisture contents of the entire hay sample in each jar. It is possible for one of the wafers from the sample to be at a higher moisture content than the others. The low moisture content samples, at times, would have one wafer with a little mold and the rest would be mold free. In this case the sample was called mold free. There

EXPLANATION OF PLATE VIII

A view of the temperature recordings from the jars. Note at bottom of figure; points 13, 14, 15, and 16 show and increase in temperature. This indicates advance stages of molding.

PLATE VIII


DEGREES FAHRENHEIT
50
75
100
125
150
DEGREES FAHRENHEIT
50
75
100
125
150
DEGREES FAHRENHEIT

EXPLANATION OF PLATE IX

A view showing the wafers in the jars. Mold is in its advanced stages.

PLATE IX

is a remote possibility that in some instances, moisture may have migrated from one end of the wafer to the other. This may be the reason for the appearance of mold on one side of the wafer only. Usually, in low moisture content wafers, mold appeared on the bottom of the wafers that were in contact with the jar. At times, the mold appeared on one side of the wafer that was also in contact with the jar surface. The other surfaces would be mold free. There is also a remote possibility that some species of mold are light sensitive and would not appear, due to the light that was left on in the room. The author on one occasion shined a powerful flood lamp on some mold that had appeared while the light in the room was off. Within a few hours, the white fuzzy material described before had disappeared.

The difference in location of the lines corresponding to the different runs on the plate are attributed to the possible difference in size of the colonies of mold present, species of mold present and environmental conditions in the jar.

Plate VII, page 27, shows the results of the time for mold to grow on the heated wafers. One sample with only 16.33% moisture molded and did not conform with what the others seemed to show as a trend. It is the author's opinion that this sample is part of the error that comes with research, and is not representative of what was generally found. Generally speaking, the results gave a curved line for each run. At times, the points were so far apart that it was very hard to say just exactly what the slopes of the lines would be. The author fitted the best lines possible for the given data.

Again, the differences in the slopes of the lines and their position on the graph can be attributed to the size of the colonies of mold present, species of mold and the differences in moisture content. It is believed

that the straighter line obtained was due to a more even distribution of the moisture within the hay sample. There is also the possibility that the heat applied during the wafering may have destroyed some of the molds that would normally appear in the moisture range of 23 to 28 percent.

The main conclusion which can be stated from the observation of these plates is that wafering hay at 20% moisture wet basis with the system described earlier in this report will produce wafers that are free from mold growth, provided the storage temperature does not go over 90° F. Conditions may change if the temperature goes higher, but should remain about the same as described by the plates. At temperatures above 130° F., mold will not grow. It can also be stated from this observation that even if the farmer were to wafer hay at higher moistures than that recommended for safe storage, he has a certain amount of time to do something about the moisture content of the wafers. For example, if a farmer were to wafer hay at 25% moisture wet basis, he would have about 70 hours before mold will grow. With an artificial drying rate of 2% per day he has almost 3 days to reduce the moisture content to a level safe for storage.

Plate X, page 36, shows the relationship of relative humidity and percent moisture content at the time mold appeared. It can be observed that generally (with a few exceptions) the wafers that did not mold never reached a relative humidity above 55%. Most molding took place in the region of 55% to 75% relative humidity. The other group that shows up is the one that extends from 75% to 93% relative humidity. Due to the time of night when the mold appeared, the author was unable to take the relative humidity reading. They were taken the next morning when moderate amounts of mold were visible. Condensation was also visible on the walls

EXPLANATION OF PLATE X

Moisture content and relative humidity at time of molding for the heated and unheated wafers.

PLATE X

% Relative Humidity
90
80
70
60
50
40
30
20
5
10
15
20
25
30
35
40
Moisture Content, % Wet Basis
o - Moderate amount of mold visible.
+ - Begining to mold.
• - No Mold.

of the jar. This obviously had a marked effect, causing the high relative humidity readings. The main conclusion from the data shown in the plate is that relative humidity is a factor in mold growth.

By observing plates XI, and XII, pages 39 and 41, the assumed equilibrium relative humidity can be observed. These data were taken after 25 hours of storage. A curvilinear and straight line regression analysis was run on the data (see Appendix for program). The author lets the reader choose the line he believes to best fit the data plotted.

The carbon dioxide results shown on plate XIII, page 43, indicated that as mold develops, the carbon dioxide content increases. The higher the moisture content, the faster the mold develops and the greater the amount of carbon dioxide released by the mold. It is the author's opinion that carbon dioxide could possibly be used as a means of an early detection of mold growth (12, 13).

The radiated wafers did show some signs of starting to mold in a similar manner to the other wafers used in the experiment. But, the mold did not continue to develop as was the case with the other wafers. After about 36 hours of little or no development, molding started again. The author believes that the start of new molding could be due to the handling of the wafers after irradiation, by mold spores in the air that settled on wafers during transfer from plastic bags to jars, or by mold spores in the air coming into the jars. With more research along these lines, radiation could in the near future be one of the answers to the mold attack on foodstuffs.

EXPLANATION OF PLATE XI

Effect of moisture content on the equilibrium relative humidity of the unheated wafers.

PLATE XI

PLATE XI

EXPLANATION OF PLATE XII

Effect of moisture content on the equilibrium relative humidity of the heated wafers.

PLATE XII

% Relative Humidity
90
80
70
60
50
40
30
20
5
10
15
20
25
30
35
40
Moisture Content, % Wet Basis

EXPLANATION OF PLATE XIII

Effect of time on the amount of carbon dioxide released by the mold. % of carbon dioxide means % by volume.

PLATE XIII

SUGGESTIONS FOR FURTHER RESEARCH

1. The fact that baled hay was used for this particular storage problem, may have had some effect on the results obtained. Therefore, the author suggests the repitition of this experiment, but with use of fresh hay, either field or laboratory cured.

2. A study on the use of inhibitors, such as Sorbic Acid or Potassium Sorbate, as a means of controlling mold growth. The author feels that the inhibitors could be mixed with the hay as the wafer is being formed. These inhibitors could possibly increase the time before mold is visible (8).

3. Further study of the possible use of carbon dioxide as a means of an early detection device. As the mold spore develops, larger quantities of carbon dioxide will be produced. A correlation of the carbon dioxide and time could be presented. With this the farmer could take samples from his storage facility and predict how long it would take for mold to develop.

4. A more thorough study should be made on the use of nuclear radiation as a means of inhibiting mold growth, and possibly sterilizing the hay. The literature indicates this to be very promising, and the results of the brief study made in this report seem to confirm it.

ACKNOWLEDGMENTS

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APPENDIX

RESULTS FOR STRAIGHT LINE REGRESSION - UNHEATED

.811194 58.664020

RESULTS FOR STRAIGHT LINE REGRESSION - HEATED

.704036 65.664181

RESULTS FOR STRAIGHT LINE REGRESSION - COMBINED DATA

.735999 62.732276

RESULTS FOR CURVILINEAR REGRESSION - UNHEATED

.296891 30.427140

RESULTS FOR CURVILINEAR REGRESSION - HEATED

.194656 44.765609

RESULTS FOR CURVILINEAR REGRESSION - COMBINED DATA

.223699 39.656627

```
C  C  STRAIGHT LINE REGRESSION - LEAST SQUARE METHOD
C     CARLOS DIAZ
C     DEPT.  AGRICULTURAL  ENGINEERING
      DIMENSION X(99),Y(99)
      READ,A
      N=A
      SUMX =0.
      SUMX2=0.
      SUMY =0.
      SUMXY=0.
      DO 47 N=1,N,1
      READ ,X(N),Y(N)
      SUMX =SUMX +X(N)
      SUMX2=SUMX2 +(X(N)**2.)
      SUMY =SUMY +Y(N)
   47 SUMXY=SUMXY +(X(N)*Y(N))
      SUM2X=SUMX**2.
      SLOPE=(SUMXY-SUMX*SUMY/A)/(SUMX2-SUM2X/A)
      B=(SUMY/A)-SLOPE*SUMX/A
      PUNCH 4,SLOPE,B
    4 FORMAT (F15.6,F15.6)
      END
```

```
C  C  CURVILINEAR REGRESSION OF A LINE - LEAST SQUARE METHOD
C     CARLOS DIAZ
C     DEPT.  AGRICULTURAL  ENGINEERING
      DIMENSION X(99),Y(99)
      READ,A
      N=A
      SUMX =0.
      SUMX2=0.
      SUMY =0.
      SUMXY=0.
      DO 47 N=1,N,1
      READ ,X(N),Y(N)
      X(N)=LOG(X(N))
      Y(N)=LOG(Y(N))
      SUMX =SUMX +X(N)
      SUMX2=SUMX2 +(X(N)**2.)
      SUMY =SUMY +Y(N)
   47 SUMXY=SUMXY +(X(N)*Y(N))
      SUM2X=SUMX**2.
      SLOPE=(SUMXY-SUMX*SUMY/A)/(SUMX2-SUM2X/A)
      b=(SUMY/A)-SLOPE*SUMX/A
      B=EXP(b)
      PUNCH 4,SLOPE,B
    4 FORMAT (F15.6,F15.6)
      END
```

DATA FOR UNHEATED HAY

18.9	70.0
18.1	67.0
17.8	73.0
19.7	68.0
24.8	83.0
25.5	76.0
25.0	78.0
24.5	83.0
25.1	83.0
26.3	85.0
27.3	87.0
25.3	87.0
33.6	85.0
33.3	85.0
36.9	83.0
33.8	83.0

DATA FOR HEATED HAY

15.3	78.0
13.6	78.0
17.6	78.0
19.5	82.0
22.9	88.0
21.5	78.0
20.1	71.0
22.1	78.0
30.7	82.0
29.3	88.0
33.3	92.0
30.8	91.0
33.6	88.0
32.8	86.0
33.0	88.0
34.5	92.0

MOLD GROWTH ON WAFERED HAY

by

JUAN CARLOS DIAZ

B. S. Agricultrual and Mechanical College of Texas, 1963

AN ABSTRACT OF A MASTER'S THESIS

submitted in partial fulfillment of the

requirements for the degree

MASTER OF SCIENCE

Department of Agricultural Engineering

Kansas State University
Manhattan, Kansas

1965

The objective of this investigation was to determine how long after a hay wafer is made and stored under certain environmental conditions, does it take for mold to appear.

Baled hay was chopped in a hammer mill, and the moisture content brought up to that needed for the experiment. The wafers were made by a closed die process. There were two types: wafers made from unheated hay, with moisture contents of 15, 20, 25, and 30% moisture wet basis, and heat lamp heated hay with moistures of 20, 25, 30, and 35% wet basis. Once out of the die the wafers were stored in gallon jars that were in a walk-in type refrigeration room in which the temperature was kept at 90°F. Air at the rate of 0.025 cubic feet per hour was pumped through the jars. Relative humidities were taken periodically. Also, some wafers were irradiated with Gamma radiation to see what effect it had on mold growth. Carbon Dioxide content samples were taken to see if some correlation existed between mold growth and carbon dioxide present.

The results show that there is a definite relationship between the moisture content of the wafers and the time for mold to grow. The higher the moisture content the faster the mold grew with the lower limit being in the range of 30 to 36 hours for moisture contents above 30% wet basis. The relative humidities varied with time all through the experiment. The assumed equilibrium relative humidities taken after 25 hours of storage, showed that as the moisture content increased, the equilibrium relative humidity also increased in a straight line relationship: $Y = 0.735(X) + 62.732$. Gamma radiation appeared to have inhibited mold growth for a period of 36 hours after which molding started. It is assumed that this molding is due to new mold spores that fell on the hay during handling or

were in the air coming into storage. Carbon dioxide tests showed that as time increases, the amount of carbon dioxide present also increases. The carbon dioxide is released by the mold spore as it attacks the plant for its food. This action varied for the moisture content available; the higher the moisture content, the faster the molding, the more carbon dioxide present.